ABSTRACT

Lysozymes are hydrolases that are divided into 6 different families, based on their distribution and function. They are c-type, g-type, i-type, plant, phage and bacterial. C-type lysozymes are predominantly expressed among a wide variety of species. Most recently, lysozyme like (LYZL) proteins that belong to the c-type lysozyme family were identified, which include the six members namely LYZL1, SLLP1/LYZL3, LYZL4, LYZL5, LYZL6 and LYZL7. Recently, the importance of mouse *SLLP1* as a receptor for glycoprotein interaction during fertilization was reported. However, the exact role of LYZL proteins in general physiology and in male reproductive function in particular has not been studied. Hence, we attempted to characterize the rat LYZL proteins with specific reference to male reproductive tract function.

Our results indicate that the rat LYZL proteins are encoded by separate genes and are not isoforms. Phylogenetic analysis showed that they are highly conserved and are distributed in various organisms. *In silico* analysis of the primary, secondary and tertiary structures of LYZL proteins revealed that they are homologous to chicken lysozyme, suggesting that *Lyzl* genes would have arose from a common ancestor. Among the LYZL proteins characterized, only LYZL1 and LYZL6 have conserved the lysozyme active site residues. *Lyzl* mRNA transcripts seem to be expressed predominantly in male reproductive tract. Their expression was not developmentally regulated and androgen independent. Immunostaining revealed the presence of these proteins in male reproductive tract tissues and on spermatozoa. LYZL1 and 6 display peptidoglycan binding property and they possess muramidase, isopeptidase and antibacterial activities, whereas the remaining proteins did not. The role of LYZL proteins in sperm function was evaluated by incubating spermatozoa with LYZL6 antibodies. *In vitro* LYZL6 neutralization seems to inhibit calcium influx and acrosome reaction suggesting that

they have important role in sperm function. *In vivo* studies using rats immunized with LYZL6 displayed spermatozoa with reduced hypermotility and decreased fertility. Taken together, these results suggests that LYZL6 may have a role in sperm function.